Amendments to the Specification:

Please amend the paragraph at page 19, lines 23-30, as follows:

In preferred embodiments, the set of parent polynucleotides can be selected according to their function. As a non-limiting example, one or more polynucleotide sequences may be identified from public sources, such as literature databases like PubMed, sequence databases like GenBank, or enzyme databases available on-line from ExPASy of the Swiss Institute of Bioinformatics, based on their ability to code for proteins capable of catalyzing a certain chemical reaction. Upon identification of a polynucleotide, other sharing homology at the nucleotide or amino acid level can be further identified using homology searching tools, such as BLAST ~~(publically available online at *www.ncbi.nlm.nih.gov/BLAST/*)~~.

Please amend the paragraph at page 20, lines 9-25 as follows:

Optimization of parent polynucleotide sets can be achieved by a variety of methods. For example, an optimized set of parent polynucleotides can be selected from a larger set of ~~polynucletides~~ polynucleotides. The basis for selection can be a specific property, function, or physical characteristic that is desirable in the recombined sequences of the library. For instance, if a recombined polynucleotide sequence capable of coding for an enzyme that catalyzes a reaction at high pH is desired, then of the possible nucleotide sequences that catalyze the reaction, only the ones that perform at high pH are selected to comprise the optimized set of polynucleotides. In another approach to making optimized sets of polynucleotides that makes fewer assumptions about the contribution of sequence to phenotype and allows greater diversity, members of the optimized set may be chosen according to phylogenies. For example, a set of polynucleotides sharing a predetermined

minimal sequence homology may be organized into a phylogenetic tree. Algorithms enabling the assembly of homologous sequences into phylogenetic trees are well known to those skilled in the art. For instance, the phylogenetic tree building program package Phylip is readily available to the public on-line ~~at *evolution.genetics.washington.edu/phylip.html* maintained by~~ through the University of Washington. Sequences representing different branches of the calculated phylogenetic tree may then be selected to comprise an optimized set of polynucleotides.

Please amend the paragraph spanning page 20, line 26 to page 21, line 12, as follows:

The set of parent polynucleotides is dissected into oligonucleotides. Oligonucleotides may be chosen randomly or based on particular features of the polynucleotides. Oligonucleotides may also be chosen in order to facilitate their coupling. For example, it may be preferable for the 5' terminus of oligonucleotides to be a C, rather than a G, because the enzyme T4 RNA ligase ligates acceptor oligonucleotides to a 5' C more efficiently. In the event the parent polynucleotides share homology, the sequences may be aligned to facilitate identification of regions of sequence appropriate to represent a subset of oligonucleotides. For instance, a highly variable or highly conserved region of sequence may be designed to represent a subset of oligonucleotides. Sequence alignments are readily performed by those skilled in the art. An example of a suitable sequence alignment program is ClustalW v. 1.7~~, available online at *clustalw.genome.ad.jp*~~. Oligonucleotides may also be designed according to size. For example, subsets having longer oligonucleotides may result in libraries with less complexity than libraries comprising shorter oligonucleotides. Furthermore, oligonucleotides not directly derived from the selected polynucleotide set can

be introduced into the library. For instance, certain mutations or degeneracies desired in the resulting library may be incorporated by adding oligonucleotides to the desired subsets (or sequence positions). Thus, great control can be maintained in engineering particular features into the library such as, but not limited to, restriction sites, point mutations, frame shifts, insertions, deletions, and the like.

Please amend the paragraph at page 21, lines 13-28, as follows:

In preferred embodiments, oligonucleotide subsets may be determined from their corresponding amino acid sequence subsets. Accordingly, in order to encode two or more amino acids at the same position in the same sequence (degeneracies), the following methods may be used. Most simply, it may be readily determined upon inspection that a basepair in one oligonucleotide differs from the analogous basepair of a further oligonucleotide, and the difference directly corresponds to a difference in one amino acid. Alternatively, a further embodiment involves determining oligonucleotide subsets from the amino acid sequences themselves. This approach may be facilitated using the computer program CyberDope which is available online ~~at *www.kairos-scientific.com/searchable/cyberdope.html*~~ and is described in Delagrave, *et al.*, *Protein Eng.*, **1993**, *supra*, Delagrave, *et al.*, *Biotechnology*, **1993**, *supra*, and Goldman, *et al.*, *supra*. According to this program, a set of amino acids, for instance occupying a variable amino acid site in a set of polypeptides, may be entered, (*e.g.*, A and S, or A, S and T). Based on the amino acids entered, the program calculates a set of degenerate codons. In alternative embodiments, the codon preferences (codon usage) of the host organism which will express the library of polynucleotides, may be taken into account when designing the oligonucleotides to avoid introducing disfavored codons.

Please amend the paragraph at page 34, lines 9-13 as follows:

The amino acid sequence of the enzyme (accession number 995625) was used to identify related sequences from the public database of sequences available online by performing a BLAST search ~~(www.ncbi.nlm.nih.gov/BLAST/)~~. Twenty-five sequences were chosen manually from 100 sequences obtained in the BLAST search results. In an alternative embodiment, the selection process may be automated.

Please amend the paragraph at page 35, lines 1-16, as follows:

The sequences of family 1 were aligned together. Without families, differences between the sequences generally limit themselves to point mutations. Such slight differences can readily be encompassed by the assembly of degenerate oligonucleotides wherein two or more bases are simultaneously encoded by the DNA synthesizer used to make the oligonucleotide. In cases where more than simple single-nucleotide differences must be encoded by a degenerate oligonucleotide, the program CyberDope~~, available online at www.kairos-scientific.com/searchable/cyberdope.html,~~ can be used to design appropriate degenerate codons. These degenerate codons are selected by the program to encode complex combinations of amino acids. Oligonucleotides, including those that are degenerate, ~~can be~~ are commercially available from companies such as Operon Inc. (Alameda, CA). Therefore, the 7 sequences of family 1 were described by 19 oligonucleotides, most of which have 2 degenerate nucleotide positions. Oligonucleotide number 13 was the most complex, encoding 7 mutations requiring 8 nucleotide degeneracies. Although the combinatorial complexity of the 19 degenerate oligonucleotides exceeds 2^{42}, or more than $4x10^{12}$ possible

sequences, the amino acid substitutions are quite conservative, such that most combinations will likely yield functional subtilisins with a variety of phenotypes.

Please amend the paragraph spanning page 35, line 29 to page 36, line 3 as follows:

Using the amino acid sequences encoded by the above ~~polynuclotides~~ polynucleotides of families 1 and 3, degenerate oligonucleotides were designed with the program CyberDope. The resulting oligonucleotides needed to synthesize the orf of families 1 and 3 are listed below. Oligonucleotides are numbered in the order in which they are to be assembled, from the 5' to the 3' end. Degeneracies are encoded according to the IUPAC code (described on p. 234 of the 2000-2001 New England Biolabs catalog or available ~~avilable, for example,~~ online at ~~*www.neb.com/neb/tech/tech_resource/miscellaneous/genetic_code.html*~~.